#### HUMANE EUTHANASIA OF BATS FOR PUBLIC HEALTH RABIES TESTING

The Michigan Rabies Working Group (RWG) is an interagency group that was formed in the late 1990's and whose members represent the Michigan Departments of Community Health, Agriculture, and Natural Resources, the United States Department of Agriculture (USDA), Michigan State University, local public health, animal control officers, wildlife enthusiasts, and animal welfare organizations. This group meets regularly to discuss rabies issues in Michigan. Providing guidance for the safe and humane euthanasia of bats for public health testing became a recent priority for the RWG. The objectives of this document include:

- Describe, in brief, the epidemiology of rabies in the U. S. and Michigan
- Discuss the public health concerns when bats are found in and around human dwellings
- Describe how to safely collect a bat for rabies testing
- Provide information about safe, humane, and effective euthanasia techniques in bats
- Provide information about packaging and shipping specimens for rabies diagnostic testing.

## About Rabies in the U.S. and Michigan

Rabies is a preventable viral disease transmitted primarily by the bite of an infected mammal. Untreated, rabies has the highest case fatality rate of any infectious disease known, being 100% fatal once symptoms begin. Any mammal can be infected with rabies, but following the implementation of rabies vaccination of domestic dogs in the 1950's, wild mammals have become the primary reservoir for this virus in the United States. Wild animals accounted for 92% of all animal rabies cases reported in 2006<sup>1</sup>. The rabies reservoir species in the U.S. include raccoons, bats, skunk, and fox. In Michigan, bats are the primary reservoir species, with a focus of skunk-strain rabies in the southeast and "thumb" area of the state. The Centers for Disease Control and Prevention has an excellent Rabies Website at <a href="http://www.cdc.gov/rabies/">http://www.cdc.gov/rabies/</a>. In addition, current information about rabies in Michigan can be found at <a href="http://www.michigan.gov/rabies">http://www.michigan.gov/rabies</a>.

## **Bats and Rabies**

It is estimated that less than 1% of the bats in the wild are infected with the rabies virus. However, on average, 4-6% of the bats that are submitted for public health testing (found in and around people's homes) to the Michigan Department of Community Health's Bureau of Laboratories (MDCH BOL) are infected. The reason for the higher rate of positive bats in public health testing is that sick bats are more likely to display unusual behavior, such as an inability to fly, and end up potentially exposing humans and unvaccinated pets to rabies. Bats can pose a particular difficulty for public health and

animal control agencies. While human rabies cases are rare in the U. S. (1-2 per year on average), the majority are caused by bat strains of rabies. In many of these cases, there has been no known contact or bite from a bat. Even seemingly insignificant bat bites can transmit rabies. The current ACIP Recommendations for Human Rabies Prevention—United States, 2008<sup>2</sup> describes a bat exposure as either direct contact with a bat, or "finding a bat in the same room as a person who might be unaware that a bite or direct contact had occurred (e.g., a deeply sleeping person awakens to find a bat in the room or an adult witnesses a bat in the room with a previously unattended child, mentally disabled person, or intoxicated person). These situations should not be considered exposures if rabies is ruled out by diagnostic testing of the bat". Thus, when a bat needs to be tested because of public health concerns, it must be euthanized in such a way as to preserve the brain for diagnostic rabies testing.

## **Collection of a Bat for Rabies Testing**

Because bats are one of the natural reservoirs of the rabies virus, it must be assumed that all bats may be infected. This presents a risk to the personnel who are involved in collecting, euthanizing and preparing rabies specimens for testing. Collection of a bat should occur in such a way as to avoid contact with the bat. If a bat is present in someone's home and you cannot rule out the possibility of exposure, do not kill or free the bat. The bat should be gently collected pending consultation with the local public health agency. There are precautions that should be taken to capture the bat safely. Tools you should have available include:

Leather work gloves Small box or coffee can Piece of cardboard Tape

Once the bat lands, wearing gloves, approach slowly and place a box or can over it. Slide the piece of cardboard under the container, trapping the bat inside. Securely tape the cardboard to the container and punch small holes (less than ½ inch in diameter) in the container for the bat to breathe. Contact your local public health or animal control agency to discuss whether the bat needs to be tested for rabies and to receive instructions on what steps need to be taken next.

## Providing for Safe, Humane Bat Euthanasia

Because any bat may be infected with rabies, all personnel handling bats for euthanasia should be vaccinated against rabies. In addition to being vaccinated, the Michigan RWG recommends that personnel involved in euthanizing bats must be properly trained in the use of common euthanasia techniques. Personnel must be provided with appropriate personal protective equipment (PPE), depending on the method of euthanasia utilized, which may include protective clothing, gloves, mask and eye protection. When using gaseous agents, care must be taken to protect personnel (proper equipment, ventilation and/or scavenging system).

The primary reference regarding humane euthanasia of animals is the "AVMA Guidelines on Euthanasia, June 2007". In addition, the American Association of Zoo Veterinarians has published "Guidelines for Euthanasia of Nondomestic Animals". While there is little objective information in the literature regarding humane techniques for euthanasia in bats, the basic principles of euthanasia apply. Death must be as painless and distress free as possible. Any technique chosen must induce loss of consciousness as quickly as possible, followed by cardiac and respiratory arrest. The technique must also be reliable, irreversible, and most importantly, safe for humans.

The AVMA Guidelines on Euthanasia classify effective euthanasia methods as "acceptable", or "conditionally acceptable". Acceptable methods are those that consistently produce a humane death when used as the sole means of anesthesia. Conditionally acceptable methods are those that by their nature might not consistently produce humane death, or are methods not well documented in the scientific literature.<sup>3</sup> A number of methods may be utilized to achieve humane euthanasia in bats. Optimal methods provide for as little contact as possible with the bat. Bats should NEVER be handled with bare hands. The following is a list of agents and methods that may be used accomplish euthanasia in bats.

## **Recommended Inhalation Agents:**

# **Acceptable Methods:**

- Inhalant anesthetics (liquid)-- ex: halothane, enflurane, isoflurane, sevoflurane, methoxyflurane, desflurane
- Carbon dioxide CO<sub>2</sub> at 70% or higher concentrations-compressed gas cylinders are the only acceptable source. (Insectivorous bats may be resistant, so this method should be paired with other methods that ensure death, such as an injectable barbiturate.<sup>4</sup>)
- Carbon monoxide CO-more dangerous to personnel, requires quality equipment

#### **Conditionally Acceptable Methods:**

- Nitrogen (N<sub>2</sub>), Argon (Ar): Not practical/available in most situations
- Ether: Should only be used in carefully controlled situations in compliance with state and federal occupational health and safety regulations<sup>3</sup>. Ether is highly flammable, explosive, and presents a potential health risk to personnel who have long-term exposure to it. Its use requires good ventilation, attention to safety (never use around a flame), sufficient exposure to assure euthanasia, and proper aeration of the carcass before storage to prevent the formation of explosive epoxides.

#### **Unacceptable Inhalation Methods:**

 Exhaust fumes from an idling gasoline internal combustion engine (unacceptable due to production of additional gases leading to inadequate concentrations of CO gas and inadequate cooling of the gas)

## **Recommended Injectable Agents:** (Both are Acceptable Methods)

- Barbiturates -intravenous or intraperitoneal (Limitation: DEA Controlled substances, must be licensed to administer)
- Potassium chloride- intravenous or intracardiac, in conjunction with an inhalation anesthetic

## **Conditionally Acceptable Physical Methods:**

- Thoracic (Cardiopulmonary, Cardiac) Compression
  - o AVMA Guidelines describe this as an alternative method for small to medium sized birds when alternative guidelines are not practical.
  - Society of Mammalogists<sup>5</sup> lists this technique for small mammals as being quick, and imparting little pain, thus meeting the criteria for euthanasia methods of the United States Department of Agriculture's Animal and Plant Health Inspection Service (APHIS).
  - While this technique is not specifically recommended for bats in the AVMA Euthanasia Guidelines, other guidelines<sup>5</sup> mention its use in small mammals. When applied properly, this technique is quick and very safe for personnel, allowing for this technique to be categorized as "conditionally acceptable".
  - This method has the advantage of being rapid, apparently painless, and preserves the brain. The technique is safe for personnel and practical for field application.
  - This technique should be restricted to use in active bats. Bats in torpor have lower respiratory rates and would take longer to succumb with this method.
  - Care should be taken to apply enough pressure to restrict respiration, but not so much as to crush the bat. The larger bats, such as Big Brown bats, will require more pressure than smaller bat species.

#### **Unacceptable Physical Methods:**

- Blunt force trauma (may destroy the brain)
- Drowning (inhumane)
- Freezing (inhumane-formation of ice crystals on the skin and in tissues may cause pain or distress. Quick freezing is acceptable in a deeply anesthetized animal only.<sup>3</sup>)
- Gunshot (may destroy the brain)

# **Examples of Recommended Euthanasia Techniques**

The overarching goal of this document is to provide for the safe, humane and effective euthanasia of bats for public health testing purposes. With that goal in mind, the following are examples of techniques that meet these criteria. These are not meant to be exhaustive. Persons who are trained and knowledgeable about euthanasia techniques may have developed their own effective methods to achieve humane euthanasia in bats. Any technique used to euthanize bats should be safe for the personnel performing the task, and produce as quick and humane a death as possible in the animal. The following techniques are provided as simple examples for those who may lack the experience with this species but are willing and able to perform this task.

# **ACCEPTABLE** EUTHANASIA METHODS FOR BATS:

- 1. CHAMBER ANESTHESIA/EUTHANASIA: A bat collected in the manner outlined above may be placed inside an anesthesia induction chamber without the need to remove the bat from the container. Ensure that there are sufficient holes in the cardboard to allow for anesthetic gas to get in. Seal and fill the chamber with an inhalation agent such as one listed above. Turn off gas and leave the bat inside the chamber to allow sufficient time for the anesthetic gas to cause euthanasia. Alternatively, the bat may be left in the chamber for sufficient time to achieve anesthesia, whereby it can be removed from the chamber and container and injected intraperitoneally with a barbiturate euthanasia solution. If CO<sub>2</sub> is used to induce anesthesia in a bat, it should be followed by an injectable euthanasia agent to assure death.
- 2. CONTAINER EUTHANASIA: A cotton ball soaked with one of the above listed liquid inhalation anesthetic gases can be placed inside a sealable heavy-duty ziplock plastic bag, along with the collection container, and left to allow sufficient time for the anesthetic gas to cause euthanasia. Alternatively, the bat may be left in the sealed bag for sufficient time to achieve anesthesia, whereby it can be removed from the bag and container and injected intraperitoneally with a barbiturate euthanasia solution.

#### **CONDITIONALLY ACCEPTABLE METHODS:**

1. USE OF ETHER (or methoxyflurane): Both of these chemicals are highly soluble in blood and induce anesthesia slowly. A cotton ball soaked with either product can be placed inside a sealable chamber, such as a heavy-duty ziplock plastic bag, along with the collection container, and left for sufficient time to cause euthanasia. **Ether is highly flammable and explosive,** and should only be used in carefully controlled situations in compliance with state and federal occupational health and safety regulations<sup>3</sup>. Good ventilation is required.

2. THORACIC COMPRESSION: The captured bat could be placed in a sealable plastic bag and/or grasped directly with a pair of tongs or hemostats, with pressure firmly applied (do not crush the bat) to the thoracic region of the bat (below the head and neck) until death occurs.

## Packaging and Shipping Specimens for Rabies diagnostic testing

At no charge, the MDCH BOL provides complete kits for submitting rabies specimens. Kits can be requested by calling (517) 335-9867 and asking for Unit #47. In addition, directions on packaging and shipping can be found on the BOL website at <a href="http://www.michigan.gov/mdch">http://www.michigan.gov/mdch</a>. Navigate to Providers/Lab Services/Specimen Submission. Acceptable specimens include whole bats or the head (or brain-including both sides of the cerebellum and brain stem) of larger mammals. Specimens should be refrigerated prior to shipping. Specimens should not be frozen. Freezing does not interfere with the accuracy of the test, but the specimen must thawed before testing can proceed, causing a delay in test completion. A Rabies Test Requisition form should be completed and included with the specimen. Shipping information is on the form.

Specimens can be delivered to the lab 24/7. Recommended methods include Fed Ex or United States Postal Service (USPS) for "overnight delivery". Specimens shipped "overnight delivery" will be delivered on the next <u>business</u> day. It is best to avoid having specimens in transit over a weekend. Specimens collected on a Friday may be refrigerated and shipped on Monday, if results are not critical. Specimens can also be delivered to the laboratory by public or private couriers or private citizens. Testing is available on the weekends by special arrangement with MDCH at 517-335-8165 Monday through Friday, and 517-335-9030 nights and weekends. Emergency testing is reserved for human exposures when timely results will prevent unnecessary post-exposure treatment. When specimens need to be delivered to the lab on a Saturday, the submitter must request "Saturday Delivery" from either Fed Ex or the USPS.

#### REFERENCES

- 1. Blanton, JD, Hanlon, CA, Rupprecht, CE. Rabies Surveillance in the United States during 2006. JAVMA, Vol 231;4:540-555.
- 2. "Human Rabies Prevention—United States, 2008. Recommendations of the Advisory Committee on Immunization Practices". MMWR 2008;57:1-28.
- 3. "AVMA Guidelines on Euthanasia (Formerly Report of the AVMA Panel on Euthanasia), June 2007", available online at <a href="http://www.avma.org/issues/animal\_welfare/euthanasia.pdf">http://www.avma.org/issues/animal\_welfare/euthanasia.pdf</a>
- 4. "Guidelines for Euthanasia of Nondomestic Animals", American Association of Zoo Veterinarians, 2006.
- 5. "Guidelines for the Capture, Handling, and Care of Mammals as Approved by the American Society of Mammalogists", Animal Care and Use Committee, Journal of Mammalogy; Nov 1998; 79,4; 1416-1431.